UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM:

Subject: EPA File Symbol/EPA Reg. No.: 4643-29
Dearcide 716

6/6/94

From: Fred Johnson Jr., Biologist
Precautionary Review Section
Registration Support Branch
Registration Division (7505W)

To: Ruth Douglas PM 32
Antimicrobial Program Branch
Registration Division (7505C)

Thru: Thomas C. Ellwanger, Section Head
Precautionary Review Section
Registration Support Branch
Registration Division (7505W)

Mary Waller T.G. for 9/23/94

Applicant: Grace, Dearborn Division
W.R. Grace & Co. - Conn.
300 Genesee Street
Lake Zurich, IL 60047-2458

FORMULATION FROM LABEL:

Active Ingredient(s):	% by wt.
Trichloro-s-triazinetrione	25%
Inert Ingredient(s):	75%
Total:	100%

BACKGROUND

Grace, Dearborn Division, has submitted acute oral, acute dermal, acute inhalation, eye irritation, dermal irritation and dermal sensitization studies to satisfy the 8-month response-product specific data request for reregistration. The product was not batched. The product is Dearcide 716 Oxidizing Microbicide, a bacteriostat and algaecide for the control of the growth of bacteria and algae in industrial and commercial recirculating water cooling towers, air washer systems and lakes, ponds and reservoirs. The active ingredient is Trichloro-s-triazinetrione. The studies were conducted at Springborn Laboratories, Inc. (SLS) and the assigned MRID Nos. are 431362-03 through 431362-08.

RECOMMENDATION

1. PRS finds the acute oral, acute dermal, acute inhalation,eye irritation and dermal studies acceptable data to support the registration of REGNO 4643-29. However, the acute inhalation study is graded Core Minimum because the MMAD for this product exceeds the Agency accepted value of 4.0 μ; actual value, 4.7 μ.

2. The dermal sensitization study is graded "Supplementary" with possibility for upgrade if positive control data conducted within six months of the submitted study test date is provided.

ACUTE TOXICITY PROFILE

Acute Oral	Category 3/G
Acute Dermal	Category 3/G
Acute Inhalation	Category 2/M
Eye Irritation	Category 1/G
Dermal irritation	Category 4/G
Dermal Sensitization	/S

LABELING

1. The signal word is "Danger".

2. The Precautionary Statements should read:

" Corrosive: Causes irreversible eye damage. May be fatal if inhaled. Harmful if swallowed or absorbed through skin. Do not get in eyes or on clothing. Do not breathe dust, vapor or spray mist. Wear goggles, face shield or safety glasses. Wear a mask or pesticide respirator jointly approved by the Mine Safety and Health Administration and the National Institute for Occupational Safety and Health. Wash thoroughly with soap and water after handling. Remove contaminated clothing and was before reuse."

3. The Statements of Practical Treatment should read:

"**If in eyes:** Hold eyelids open and flush with a steady, gentle stream of water for 15 minutes.Get medical attention"

"**If inhaled:** Remove victim to fresh air. If not breathing, artificial respiration, preferably mouth to mouth. Get medical attention."

"**If swallowed:** Drink promptly a large quantity of milk, egg white, gelatin solution, or if these are not available, large quantities of water. Avoid alcohol. Get medical attention. Note to Physician: Probable mucosal damage may contraindicate the use of gastric lavage."

"**If on skin:** Wash with plenty of soap and water. Get medical attention."

4. Additional labeling may be required upon submission of outstanding information.

5. Add the following "Note to Physician":

NOTE TO PHYSICIAN: Probable mucosal damage may contraindicate the use of gastric lavage.

DATA REVIEW FOR ACUTE ORAL TOXICITY TESTING (§ 81-1)

Product Manager: (32)
MRID No.:431362-03

Reviewer: F.Johnson
Report Date: 12/21/93
Report No.: 3324.1

Testing Facility: Spingborn Labs., Inc.
Authors: Kimberly L. Bonnette, M.S., LATG

Quality Assurance (40 CFR §160.12): Attached

Test Material: Dearcide 716

Species: Sprague-Dawley Crl:CD®BR VAF/Plus® rats
Age: Young adult
Sex: 5 males and 5 females/ dose, 4 doses
Weight: (M) 213-301; (F) 217-283 g
Source: Charles River Laboratories, Inc. Portage, Michigan

Conclusion:

1. LD_{50} (mg/kg) Males = 3195 (2615-3903)mg/Kg
   Females = 2658 (2352-3003)mg/Kg
   Combined = 2947 (2634-3297)mg/Kg
2. The estimated LD_{50} is
3. **Tox. Category:** III **Classification:** Guideline

Procedure (§81-1): The test article, described as a white powder, was prepared for dosing, as a suspension, by grinding in a mortar and passing through a No. 40 mesh and mixing with Mazola Corn Oil prior to dispensation. The dosing suspension was continuously stirred during dosing. The animals chosen for the study were weighed and fasted overnight. On day 0, the test article was administered orally by gavage, individual doses calculated based on the basis of their fasted weights. Study animals were observed for clinical abnormalities at least twice on day 0 (postdose) and daily thereafter for the duration of the 14 day study. Each day a mortality check, morning and night, was made. Individual animal body weights were recorded on days 0, 7 and 14. Gross necropsies were performed on all animals, those dying during the study and those euthanized (by carbon dioxide inhalation) at the end of the study. The only deviation reported during the study is the relative humidity, reported as 51-92% rather than 40-70%, not perceived as having had an adverse effect on the outcome of the study.

Results:

Dosage (mg/kg)	(Number Killed/Number Tested)		
	Males	Females	Combined
2000	0/5	0/5	0/10
2800	---	3/5	3/5
3500	3/5	5/5	8/10
5000	5/5	5/5	10/10

Observations: As recorded above, 8/15 (53.3%) of males and 13/20 (65%) of females died during the study. In life clinical observations included salivation, decreased activity, wobbly gait, shallow breathing, reduced feces, fecal and urine staining, with dehydration and emaciation in both males and females. Additionally, females exhibited some piloerection and darkness around the eyes.

Gross Necropsy: Some observations at necropsy of those animals, both males and females, dying during the study included congested meningeal vessels in the brain, small intestines with reddish-yellow, yellowish-white and greenish-yellow mucoid contents. Some additional findings in both sexes included stomach contents of clear colorless fluid, yellowish-white granular and yellowish-white creamy material, thymus with dark red foci and hair coats with either wet clear colorless matting or wet yellowish matting. Those animals necropsied, following euthanasia, showed few remarkable findings.

DATA REVIEW FOR ACUTE DERMAL TOXICITY TESTING (§81-2)

Product Manager: (32) **Reviewer:** F. Johnson
MRID No.:431362-04 **Report Date:** 11/23/93
Report No.: 3324.2

Testing Laboratory: Springborn Labs., Inc.
Author(s): Kimberly L. Bonnett, M.S., LATG
Quality Assurance (40 CFR §160.12): Attached
Test Material: Dearcide 716
Species: New Zealand White rabbits
Weight: (M) 2131-2193; (F) 1980-2226 g
Age: Adult
Source: Mohican Valley Rabbitry, Loudonville, OH

Summary:

1. LD_{50} (mg/kg): Males =
   Females =
   Combined =

2. The estimated LD_{50} is > 2000 mg/Kg

3. **Tox. Category:** III **Classification:** Guideline

Procedure (§81-2): This study was conducted as a single dose, " Limit Test" using ten New Zealand White rabbits (five males and five females). The test article was administered as received, as a white powder. On the day prior to dosing, the animals were prepared for dosing by removing the fur from the dorsal trunk area using an animal clipper; the clipped area was approximately 10% of the animal's body surface (BSA). The BSA was calculated for each animal using the formula: [BSA in cm^2= 9 x (body weight in grams)$^{0.66667}$] and the four corners of this area were delineated on an appropriately sized 4 ply porous gauze dressing. The test site was moistened to enhance test article contact with the skin by wiping the site with gauze moistened in distilled water. Test article was then spread evenly over the delineated area of the gauze dressing. The gauze dressing, backed by plastic wrap (occlusive binding) was applied to the clipped area of the animal and the elastic wrap secured around the trunk and test area.The elastic wrap was further secured with adhesive tape around the trunk at the cranial and caudal ends. Individual doses were calculated on the basis of animal's day 0 body weight. After dosing, collars were placed on animals and remained in place for the duration of the 14 day study. After the approximate 24-hour exposure period, the gauze dressing, plastic and elastic wrap were removed, the corners of the test site delineated using a marker and residual test article removed using gauze moistened with distilled water. Animals were observed for clinical abnormalities, twice on the day of dosing and daily thereafter throughout the study, while examinations for erythema and edema were made following patch removal and daily thereafter through

day 14. Individual animal body weights were recorded prior to dosing, on day 0 and on days 7 and 14. Following euthanization of all test animals at the end of the 14 day study period, gross necropsies were performed on each. It is reported that 1/5 males lost both the patch and binding during dosing; the test site was wiped with distilled water and a new patch and binding was placed in the manner earlier described. Additionally, 1/5 males was replaced following dosing; developed diarrhea during dosing. The test article was not exposed to the skin surface consistent with guidelines; test article should be dissolved or suspended in the vehicle and evenly applied to the skin, not to the patch.

Results:

Reported Mortality

DOSAGE (mg/Kg)	(NUMBER KILLED/NUMBER TESTED)		
	Males	Females	Combined
2000	0/5	0/5	0/10

Signs of toxicity: Positive clinical findings included 10/10 with irritation outside of the test site, erythema with blanching in 10/10 animals (10-50%), eschar-exfoliation 10/10 animals and grade 3-4 edema in 10/10 animals. No other clinical findings are reported.

Gross Necropsy Findings: Other than 2/5 females with reported oviduct(s) cysts filled with clear fluid and measuring 0.3-0.4 cm in diameter, and gross thickening of treated skin, no other significant findings are reported.

DATA REVIEW FOR ACUTE INHALATION TOXICITY TESTING (§81-3)

Product Manager: (32) **Reviewer:** F. Johnson
MRID No.: 431362-05 **Report Date:** 02/04/93
Report No.: 3324.3

Testing Laboratory: Springborn Labs., Inc. (SLS)
Author(s): Rusty E. Rush, M.S., LAT
Quality Assurance (40 CFR §160.12): Attached
Test Material: Dearcide 716
Concentration:
Species: Sprague-Dawley Crl:CD
Weight: (M) 226 - 284 g; (F) 251 - 257 g

Age: Young Adult
Sex: 5 males and 5 females/dose; 3 doses
Source: Charles River Laboratories, Inc., Portage, Michigan

Summary:

1. LC_{50} (mg/kg): **Males** = 0.468 (0.170 - 1.250) mg/L
Females = 0.766 (0.282 - 2.081)mg/L
Combined = 0.572 (0.361 - 1.034) mg/L
2. **The estimated LC_{50} is**
3. **MMAD:** 4.7 μ
4. **Tox. Category:** II **Classification:** Core Minimum

Procedure (§81-3): Prior to the conduct of this study, attempts at particle size reduction by hammer and ball milling procedures were undertaken Ricerca, Inc. Upon receipt by the testing laboratory, the test article was placed in a 40-43°C drying oven in order to maintain a dry test article and to reduce agglomeration. The test article was passed through a flour sifter prior to the aerosol generation procedure as an attempt to reduce any particle agglomeration that may have occurred subsequent to the hammer and air milling procedure. These procedures were performed in order to reduce the test articles' physical and aerodynamic particle size prior to aerosol generation. Details of the procedures used for particle size reduction are submitted with this submission as MRID No. 431362-01 (Document No.: 5822-93-0221-FO-001). Preliminary aerosol generation trials were performed and are recorded. The test aerosol was generated with an NBS dust aerosol generator. On day 0, animals were weighed and placed in the 100 liter whole-body inhalation chamber and dosed at aerosolized test article doses of 0.052, 0.52 and 5.41 mg/L. Aerosol concentration was measured in the chamber by the gravimetric method. Chamber temperature, and humidity were measured electronically and recorded at approximately 30 minute intervals. Aerodynamic particle size distribution was determined twice during each aerosol exposure by a 7 L/min. cascade impactor providing the data to compute the MMAD and GSD. Chamber oxygen content was also measured and recorded at 30 minute intervals.

Upon removal of animals from exposure chamber,attempts were to remove residual test article from animal's coats by vacuuming, with tap water and towel-drying. Individual animal body weights were recorded before dosing on day 0 and days 7 and 14. Observations were made clinical abnormalities on days 0 (twice/day), 7 and 14. Mortality checks were made twice daily. Gross necropsy examinations were performed on all animals dying spontaneously and those surviving the study (euthanized by carbon dioxide inhalation).

Results:

Reported Mortality:

Exposure Concentration mg/L	(NUMBER KILLED/NUMBER TESTED)		
	Males	Females	Combined
0.052	0/5	0/5	0/10
0.52	3/5	1/5	4/10
5.14	5/5	5/5	10/10

mg/l

Nom. Conc.	Grav. Conc.	Analyt. Conc.	MMAD	GSD	Temp[F]	Hum%	Air Flow [l/m]
0.48	0.052	----	3.8μ	1.5	69.6	30.95	300.2
2.65	0.52	----	4.7μ	2.4	73.7	31.0	379.5
12.08	5.14	----	4.8μ	1.9	76.0	40.38	410.6

Clinical Observations: Some in life clinical observations include labored breathing, gasping, wobbly gait, activity decrease, piloerection, hair loss, salivation and urine stain.

Gross Necropsy Findings: Remarkable necropsy findings in animals dying during the study mottled or consolidated lungs, yellowish-tan mucoid contents in the small intestines, enlarged cervical lymph node (2/5 females and 1/5 males dosed at 5.14 mg/Kg) and hair coats with wet white matting.

DATA REVIEW FOR PRIMARY EYE IRRITATION TESTING (§81-4)

Product Manager: (32)
MRID No.:431362-07

Reviewer: F. Johnson
ReportDate:11/23/93
Report No. :3324.4

Testing Laboratory: Springborn Labs.Inc.,(SLS)
Author(s): Kimberly L. Bonnette, M.S. LATG

Quality Assurance (40 CFR §160.12): Attached

Test Material: Dearcide 716
Dosage: 0.10 g
Species: New Zealand White rabbit
Sex: One male
Weight: Not provided
Age: Adult
Source: Mohican Valley Rabbitry, Loudonville, Ohio

Summary:

1. Toxicity Category: I

2. Classification: Guideline

Procedure (§81-4): Animals received pre-test ocular screen; examined macroscopically with the aid of an auxiliary light source. In addition, the corneal surface was examined using fluorescein sodium dye; retention of dye using a long-wave UV light source. The test article was prepared for dosing by being ground and passed through a No. 40 mesh sieve. Dosing consisted of 0.1 g of test article instilled into the conjunctival sac of the right eye of one animal, only, after gently pulling the lower lid away from the eye. Following instillation, the eyelids were gently held together for approximately one second. The contralateral remained untreated to serve as a control.Because of the observed severe irritation, only a single one hour observation was made; both eyes were examined and the animal was euthanized (intravenous injection of sodium pentobarbital). The relative humidity of the animal room (78%) is reported as a protocol deviation, but not believed to have had an adverse effect on the outcome of this study.

Results:

Observations	(number "positive"/number tested)							
	Hours	Days						
	1	1	2	3	4	7	14	21
Cornea Opacity	1/1	-	-	-	-	-	-	-
Iris	1/1	-	-	-	-	-	-	-
Conjunctivae								
Redness	1/1	-	-	-	-	-	-	-
Chemosis	1/1	-	-	-	-	-	-	-
Discharge	1/1	-	-	-	-	-	-	-

Comments: Study discontinued after the first hour observation of the one animal dosed because of severe corneal opacity. The test article is being placed in Category I for eye irritation.

DATA REVIEW FOR SKIN IRRITATION TESTING (§81-5)

Product Manager: (32) **Reviewer:** F. Johnson
MRID No.: 431362-08 **Report Date:** 11/23/93
Report No.: 3324.5
Testing Laboratory: Springborn Labs., Inc.
Author(s): Kimberly L. Bonnette, M.S., LATG

Quality Assurance (40 CFR §160.12): Attached

Test Material: Dearcide 716
Dosage: 0.5 g
Species: New Zealand White rabbit
Age: Adult
Sex: 6 females
Weight: 2.0 -3.5 Kg
Source: Mohican Valley Rabbitry, Loudonvliie, Ohio

Summary:

1. **Toxicity Category:** IV

2. **Classification:** Guideline

Procedure (§81-5): On day 1 the test animals had the fur removed from the dorsal area of the trunk using an animal clipper, care being taken to avoid abrading the skin during the clipping process. On the following day (day 0), the test article was applied to a small area of intact skin on each animal (approximately 1"x 1"). Prior to application of the test article, 0.5 g, the test sites were moistened with gauze moistened in distilled water. The gauze was placed over the test article; an elastic wrap was placed over the trunk and test area (semi-occlusive binding). The entire gauze and wrapped area were further secured with adhesive tape around the trunk at the cranial and caudal ends. Collars were placed on each animal following dosing, and remained in place until day 3. The 4 hour exposure period was followed by removal of all patches and wrappings; residual test article was removed using gauze moistened with distilled water. Animals were examined for signs of erythema and edema at 1, 24, 48 and 72 hours and on day 7 following patch removal. The dermal test sites were reclipped as necessary to allow visualization of the skin. Clinical observations made and any unusual findings were recorded; mortality checks were performed twice daily. Reported protocol deviations include room temperature and humidity excesses, (68-73°F and 63-80%) rather than (61-70°F and 40-60%). These deviations are not considered to have had an adverse effect on the outcome of this study. The test article was not exposed to the skin surface consistent with guidelines; test article should be dissolved or suspended in the vehicle and evenly applied to the skin, rather than overlaying the test site with a patch to which the test substance has been applied.

Results: Exposure to the test article produced very slight to well defined erythema and very slight edema at 24 hours with very slight edema by the 48 and hour scorings. All dermal irritation had abated by study day 7.

Special Comments:

DATA REVIEW FOR DERMAL SENSITIZATION TESTING (§81-6)

Product Manager: (32) **Reviewer:** F. Johnson
MRID No.: 431362-04 **Report Date:** 12/17/93
Report No.: 3324.6

Testing Laboratory: Springborn Labs., Inc.
Author(s): Kimberly L. Bonnette, M.S., LATG
Quality Assurance (40 CFR §160.12): Attached
Test Material: Dearcide 716
Positive Control Material: 1-chloro-2,4-dinitrobenzene (DNCB)
Species: Hartley-derived albino guinea pig
Weight: Induction, (M) 338-392 g; (F) 367-446 g
Challenge Control (M) 336-392 g; (F) 359-374 g
Rechallenge Control (M) 337-385 g; (F) 365- 394 g
Age: Young adult
Source: Harlan Sprague Dawley, Inc., Indianapolis,Indiana

Method: Modified Buehler

Summary:

1. **This Product is / is not a dermal sensitizer.**

2. **Classification:** Supplementary

Procedure (§81-6): Topical range-finding studies were conducted on Dearcide 716 at concentrations of 100%, 75%, 50%, 25%, 10%, 5%, 2.5%, 1.0%, 0.5%, 0.2% and 0.1% in mineral oil. The 100% concentration of test article was chosen as the dose for induction. On the day prior to dose administration, the hair was removed from the left side of the animals with a small animal clipper, care being taken to avoid abrading the skin. The mode of administration of test article is described as follows:
" the test article was applied as a powder; test sites moistened with gauze in distilled water prior to application of the Hilltop Chamber". Following patch/chamber application, the trunk of the animal was wrapped with elastic wrap and secured with adhesive tape. Approximately six hours after patch/chamber application, the elastic wrap tape and patches/chambers were removed . The test sites were then wiped with gauze moistened in distilled water for induction 1 and in mineral oil then a dry gauze patch to remove residual test article for inductions 2 and 3. The three induction applications were followed by a two week rest period, followed by a challenge application, test article concentration at 0.2%, of six hours to each animal, in a manner similar to that described for induction, except that test site was on the right side. Following patch and elastic wrap removal, the test sites were wiped with gauze moistened with mineral oil to remove residual test material. Six days following the challenge application, a rechallenge was performed, test article concentration at 0.05%. The vehicle used for each induction, challenge and rechallenge was mineral oil and a vehicle control is included in the study. Observations for irritation

were made at 24 and 48 hours, Positive control data is presented as historical data using 1-chloro-2,4- dinitrobenzene (DNCB). A dermal irritation grading scale is included.

Results: Severe erythema with edema are reported at the first induction, both at 24 and 48 hours using the test article at 100% concentration. At the second induction, concentration reduced to 25%, 5/10 animals again show severe erythema with edema while 5/10 show no reaction to slight patchy erythema to slight but confluent/moderate and/or no to slight edema. Following the third induction, at a test article concentration of 25%, at 24 and 48 hours there severe erythema with edema, including blanching and eschar, reported in 9/10 animals. At challenge, at both 24 and 48 hours, no erythema to slight patchy erythema with no edema are reported. Similarly, the rechallenge shows no significant irritation as do the naive control animals for both the challenge and rechallenge.

ACUTE TOX ONE-LINER

1. PC CODE: 081405
2. CURRENT DATE: 06/02/94
3. TEST MATERIAL: Dearcide 716

Trichloro-s-triazinetrione 25%

Study/Species/Lab/ Study#/Date	MRID No.	Results	Tox. Cat.	Core Grade
Acute oral/ rat/ Springborn Labs., Inc./3324.1/ 12-21-93	431362-03	LD_{50}:Males = 3195 (2615-3903) mg/Kg Females = 2658 (2352-3003) mg/Kg Combined = 2947(2634-3297) mg/Kg	III	G
Acute dermal/ rabbit/ Springborn Labs., Inc./ 3324.2/ 01-23-93	431362-06	LD_{50} is >2000 mg/Kg	III	G
Acute inhalation/rat/ Springborn Labs., Inc./ 3324.3/ 02-04-93	431362-05	LD_{50}: Males = 0.460 (0.170-1.250) mg/L Females = 0.766 (0.282-2.081) mg/L Combined = 0.572 (0.361-1.034) mg/L	II	M
Eye irritation / rabbit/Springborn Labs.,Inc./ 3324.4/ 11-23-93	431362-07	Test article is a severe irritant; study discontinued after 1 hr.	I	G
Primary dermal irritation/rabbit/ Springborn Labs.,Inc./ 3324.5/ 11-23-93	431362-08	Very slight erythema and edema at 72 hours	IV	G
Dermal sensitization/ guinea pig/ Springborn Labs., Inc./3324.6 12-17-93	431362-04	----------	---	S

Core Grade Key:
G = Guideline
M = Minimum
S = Supplementary